IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant:	Noble et al.	Examiner:	Lundgren, Jeffrey S.
Serial No.:	10/563,077	Group Art Unit:	1639
Filed:	December 29, 2005	Docket:	UCLA.154-US-WO
Title:	GENETIC MARKER OF RESPONSE TO ATYPICAL ANTIPSYCHOTICS AND ANTIDEPRESSANTS AND METHODS FOR USE THEREOF		

DECLARATION OF DR. ERNEST P. NOBLE UNDER 37 C.F.R. §1.132

I, ERNEST P. NOBLE, declare as follows:

1. I am a named inventor on the patent application identified above, and am authorized by the Assignees to make this declaration.

2. I am Professor of Psychiatry & Biobehavioral Sciences, Emeritus, Director of the Alcohol Research Center, and Member of the Brain Research Institute at the University of California Los Angeles (UCLA). After many years as a faculty member at Stanford University and the University of California at Irvine, I became the Director of the National Institute on Alcohol Abuse and Alcoholism (NIAAA) in Washington, DC (1976-78). After coming to UCLA in 1981, I was appointed the Pike Professor of Alcohol Studies and Director of the UCLA Alcohol Research Center. I was also former Vice-President of the National Council on Alcoholism, and former President of the International Commission for the Prevention of Alcoholism and Drug Dependency. I received a B.S. degree in Chemistry from the University of California at Berkeley, a Ph.D. in biochemistry from Oregon State University, completed a postdoctoral fellowship and an M.D. at Case Western Reserve University, followed by medical internship and psychiatric residency training at Stanford University. I performed research as a Fulbright Scholar at the Sorbonne in Paris, France, as a Guggenheim Fellow at the Centre de Neurochimie in Strasbourg, France, and as a Senior Fulbright Scholar at the Max-Planck Institute for Psychiatry in Munich, Germany. I have authored over 400 scientific and medical publications and am a member of numerous editorial boards and scientific and professional societies, and have received many honors.

3. I have reviewed the above-identified patent application and the Office Action dated October 29, 2009, issued in connection with the above-identified patent application.

4. The data presented in the working examples of the above-identified patent application show that patients exhibiting the Taq1A allele (A1+ genotype) of the D2 dopamine receptor (DRD2) gene are candidates for treatment with low dose, low DRD2 binding atypical antipsychotic medications and/or selective serotonin receptor inhibitors (SSRIs), while patients lacking the Taq1A allele (A1- genotype) are candidates for treatment with high dose or high binding antipsychotics or alternative antidepressant. Contrary to the assertion at page 4 of the Office Action dated October 29, 2009, the experimental data described in the above-identified patent application supporting this statement are not vague. Analysis of the experimental results presented in the working examples of the above-identified patent application clearly conveys that the statements at paragraph 0006 of the specification are in error, as the cited statements are directly opposite of what one would glean from the data. The erroneous nature of the statements in the specification in paragraph 0006 at page 2, lines 12-17, is readily apparent because the experimental data show that A1+ patients, and not A1- patients experience more adverse effects with high doses of atypical antipsychotics or when treated with high DRD2 binding antipsychotics. Thus, it would make no sense to treat A1+ patients with high dose high DRD2 binding atypical antipsychotics as recited in lines 2-3 of claim 2 of the application as originally filed. The data also show that treatment with SSRIs is beneficial for A1+ patients, but not for A1- patients, confirming that the latter part of this phrase ("and/or SSRIs") in claim 2 as originally filed is correct.

5. Treatment of psychiatric patients with antipsychotic and antidepressant medications yields varying results with individual patients. Hyperprolactinemia is a common problem for patients treated with antipsychotic medications. The patent application states that "although use of tighter binding agents is generally associated with higher prolactin levels it is a common clinical observation that there are considerable individual variations in prolactin levels induced by identical medication at a given dose." (See para. 0050 at p. 16, l. 14-16.) Likewise, variable outcomes have been observed when treating post traumatic stress disorder (PTSD) with SSRIs. As described in paragraph 0071 at p. 26 of the patent application, a variety of studies have shown response rates approximately 20% greater than placebo, and 30-50% of patients do not respond to SSRI medication. Thus, the invention addresses the need for predictors of

response to antipsychotic and antidepressant medications. The objective for the psychiatric treatment is to minimize hyperprolactinemia in response to antipsychotic medication, and to avoid administering SSRIs to those patients who will not respond to this class of medication.

6. The problem of identifying which patients should be treated with which antipsychotic medications is addressed by the experimental data presented in Example 2, at pages 14-24 of the specification. Our work has shown that A1+ patients, as compared to A1- patients, "have significantly higher prolactin levels when treated with a variety of antipsychotic medications", and this allelic difference in prolactin response is most dramatic for patients treated with the loose binding agent Clozapine (see Table 1 at page 21, and paragraph 0063 at page 22, lines 3-4). As noted in paragraph 0060 at page 20, lines 20-23, "when all the antipsychotics were considered together patients carrying the A1+ allele had a significant and about a 40% higher prolactin levels than patients carrying the A1- allele ($F(1,142) = 4.50$, $P = .036$)." As described in paragraph 0061 at page 21, a chi-square analysis comparing allelic status across the groups with prolactin levels in the normal range and those with hyperprolactinemia was significant ($\chi 2(1)=5.523$, $p=0.018$). When analyzed in this manner, looking at number of patients falling into the hyperprolactinemia category, only 5% of the patients taking the low binding agents Clozapine and Olanzapine suffered from hyperprolactinemia, compared to 81% of those taking the high binding agent Risperidone. These data show that "optimal therapeutic effect is likely to be obtained at lower doses in A1+ schizophrenics. A1- patients may require a higher dose for maximal antipsychotic effect" (page 24, lines. 6-8). This observation that atypical antipsychotics, including Clozapine, raise serum prolactin levels (especially in A1+ patients) runs counter to the hypothesis of Kapur & Seeman (2001, Am. J. Psychiatry 158:3, see "Conclusions" portion of abstract, copy provided with Amendment of October 6, 2008) that the fast dissociation of atypical antipsychotics from the D2 receptor permits an antipsychotic effect without prolactin elevation and other adverse side effects.

7. The data described in the preceding paragraph were analyzed using techniques, analysis of variance and Yates corrected chi-square, that take into account multiple variables (genotype, gender, medication). This type of analysis is particularly important when evaluating prolactin levels, as normal prolactin levels differ for males and females. As noted in paragraph 0061 of the specification, patients in this study were classified based on whether their prolactin levels were within the normal range (based on

community sample cut-off levels set at a 95% reference range, 430 U/l for men and 560 U/l for women) or above this level (referred to as hyperprolactinemia, in this analysis). The F and $\chi 2(1)$ values reported are thus much more informative than the raw means presented in Table 1 at page 21 of the specification. The results showed that 81% (40 of 49) of patients treated with the high binding agent Risperidone exhibited hyperprolactinemia, while only 7 of 62 patients treated with the low binding agents Clozapine and Olanzapine exhibited hyperprolactinemia. When allelelic status of the patients in the group with prolactin levels in the normal range was compared to allelic status of the group with hyperprolactinemia, a significant difference was found ($\chi 2(1)=5.52$, $p=0.02$, as rounded off in the published version of this study, copy submitted herewith). Thus, the data provide conclusive and unambiguous support for treating A1+ patients with low doses of low binding atypical antipsychotics in order to minimize the risk of hyperprolactinemia. A1- patients are more able to manage treatment with either higher doses or higher binding antipsychotic medications without adverse side effects.

8. Further support for not treating A1+ patients with high DRD2 binding antipsychotic medications is provided by the data presented in Example 1 at pages 8-14 of the above-identified patent application. Example 1 shows that allelic status of the DRD2 gene is associated with another adverse effect of antipsychotic medication, extrapyramidal syndrome (EPS). Even with low doses of Risperidone, some patients experience EPS (specification at page 8, line 18, to page 9, line 2). Example 1 compared patients receiving 2-3 mg/day Risperidone (low dose) with those receiving 4-6 mg/day (high dose) and took into account A1+ or A1- allelic status. The results showed a significant gene by dose interaction ($p=0.028$), as A1+ patients fared poorly (high EPS scores) with low doses of this tight binding agent. These data were analyzed using univariate analysis of variance (ANOVA) to test between-subject effects (see paragraph 0039 at page 11 of the application). The results are summarized in paragraph 0040 at page 12, and show that, at high doses of this high DRD2 binding antipsychotic, A1- ("A2") patients had worse symptoms than A1+ patients (df 1,23, $F=4.790$, $p=0.039$), while at low doses, there was a trend for A1+ patients to have more severe symptoms (df 1,21, $F=3.4$, $p=0.08$). The data presented in Example 1, therefore, support the use of low doses of the high DRD2 binding antipsychotic Risperidone with A1- patients.

9. The data presented in Example 3 at pages 25-35 of the above-identified patent application address the problem of identifying which patients will respond well to

treatment with SSRIs. The results show that allelic status of the DRD2 gene differentiates PTSD patients who respond well to treatment with the SSRI paroxetine from those who do not benefit from this medication. Patients (63 war veterans; 2 subjects were dropped from the study from the 65 noted in the application) were assessed at baseline and again after 8 weeks of treatment with paroxetine, using the General Health Questionnaire-28 (GHQ). The results of this study are detailed at pages 30-32 of the application and summarized in the following paragraphs. As indicated in paragraph 0083 at page 30, the distribution of genotypes in the study (26 A1+ and 39 A1-) did not deviate from Hardy-Weinberg equilibrium (χ^2=2.36, P=0.091), and there was no significant difference in the ages of A1+ and A1- subjects. In the final analysis for the published version of this study, there were 25 A1+ and 38 A1- patients, and the Hardy-Weinberg equilibrium showed χ^2=2.67 (P=0.012). After taking into account the subjects who discontinued the study, the remaining subjects did not differ in age across A1+ and A1- subjects, and they did not differ in baseline total GHQ score from those who dropped out of the study. The statistical analyses used are described in paragraph 0082 at page 30 of the application, which notes that *p* values of 0.05 or less were considered statistically significant. In all bar graphs presented in these studies, the error bars indicate the standard deviation from the mean.

10. Figure 4 of the application is a bar graph showing the baseline GHQ scores, including the total score and each of the 4 subscale scores, for A1+ and A1- patients. Paragraph 0011 at page 3 provides the *P* values for the statistically significant differences observed between A1+ and A1- patients in the GHQ total score (0.040), anxiety/insomnia (0.046), social dysfunction (0.033), and depression (0.011). This figure establishes that A1+ patients were suffering more symptoms at the outset of treatment than the A1- patients. Figure 5 shows a bar graph of the baseline and post-treatment GHQ total scores for all subjects combined and for A1+ and A1- patients viewed separately. This shows that, although a statistically significant effect of treatment was observed when analyzing total scores for all subjects combined (P=0.014), when GHQ total scores are separated by allelic status, the greater difference observed for A1+ patients is not statistically significant, and neither is the lesser difference observed before and after treatment in A1- patients. However, when the subscale scores are analyzed separately, as shown in Figure 6, a statistically significant (P=0.026) difference appears between baseline and treatment scores for the social dysfunction subscale in A1+ but not in A1- patients. Differences in the mean scores for the subscales for

anxiety/insomnia and depression are apparent, but do not reach statistical significance. Given the statistically significant difference for "total patients" and the very small difference in A1- patients in these two subscores, it appears likely that a larger sample size would result in a statistically significant difference in these two subscale scores for A1+ patients as well.

11. As described in paragraph 0090 at page 32 of the application, analysis of variance in the GHQ total and subscale scores at baseline and at the end of paroxetine treatment showed no significant differences between A1+ and A1- patients: GHQ total ($F(1,43) = 0.001$, $P = 0.99$); GHQ1 ($F(1,43) = 1.5$, $P = 0.23$); GHQ2 ($F(1,43) = 0.14$, $P = 0.71$); GHQ3 ($F(1,43) = 1.1$, $P = 0.30$); GHQ4 ($F(1,43) = 2.5$, $P = 0.12$). Thus, the significant difference in GHQ scores based on allelic status observed at baseline (shown in Figure 4) disappears following paroxetine treatment (see Figure 6). Taken as a whole, the results presented in this study show that A1- PTSD patients do not benefit from SSRI treatment, while A1+ PTSD patients do exhibit significant improvement at least in social dysfunction, and likely in anxiety/insomnia and depression as well.

12. The studies described in the examples of the above-identified patent application were published in peer-reviewed journals (see Lawford et al., *Eur Neuropsychoparmacol.* 2003 13(5):313-20; Young et al. *Br J Psychiatry* 2004 185:147-51; and Lawford et al. *Eur Psychiatry* 2006 21(3):180-5). These articles have been well-received and often cited. One example of a later article that both cites our work and confirms the results relating to susceptibility to hyperprolactinemia in response to antipsychotic medication as a function of Taq1A allelic status is Calarge et al. *Pharmacogenomics and Genomics* 2009 19:373-382 (copy submitted herewith).

13. I further declare that all statements made herein of my own knowledge are true and all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both under Section 1001 of Title 18

of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issuing thereon.

Date: 3/29/10

Ernest P. Noble

Ernest P. Noble, M.D., Ph.D.